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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 :
C07K 14/725, A61K 38/17, 45/05, C07H 21/00, G01N 33/53

A1

(11) International Publication Number: **WO 96/11214**

(43) International Publication Date: 18 April 1996 (18.04.96)

(21) International Application Number: PCT/US95/12686

(22) International Filing Date: 10 October 1995 (10.10.95)

(30) Priority Data:
111196 7 October 1994 (07.10.94) IL

(60) Parent Application or Grant
(63) Related by Continuation
US Not furnished (CIP)
Filed on Not furnished

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(81) Designated States: AM, AT, AU, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LT, LU, LV, MD, MG, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TT, UA, UG, US, UZ, VN, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG), ARIPO patent (KE, MW, SD, SZ, UG).

Published
With international search report.

(54) Title: PEPTIDES AND PHARMACEUTICAL COMPOSITIONS COMPRISING THEM

(57) Abstract

The present invention relates generally to peptide sequences, and methods of their use, which sequences modulate the activity of anti-idiotypic T cells. The activity of the anti-idiotypic T cells of interest is related to the ability of these T cells to recognize anti-p277 T cells. The peptides of the present invention thus comprise important tools in the effort to diagnose, prevent, alleviate or treat disease related to insulin-dependent diabetes mellitus (IDDM).

PEPTIDES AND PHARMACEUTICAL COMPOSITIONS COMPRISING THEM

Field of the Invention

The present invention relates generally to peptide sequences, and methods of their use, which sequences modulate the activity of anti-idiotypic T cells. The activity of the anti-idiotypic T cells of interest is related to the ability of these T cells to recognize anti-p277 T cells. The peptides of the present invention thus comprise important tools in the effort to diagnose, prevent, alleviate or treat disease related to insulin-dependent diabetes mellitus (IDDM).

Background of the Invention

Type I diabetes, or IDDM, is an autoimmune disease caused by T cells that attack and destroy the insulin-producing cells located in the islets of the pancreas (23). The autoimmune process culminating in IDDM begins and progresses without symptoms. The disease surfaces clinically only when the cumulative loss of β-cells exceeds the capacity of the residual β-cells to supply insulin. Indeed, the collapse of glucose homeostasis and clinical IDDM is thought to occur only after 80-90% of the β-cells have been inactivated by the immune system. Thus, patients who can be identified as suffering from IDDM are bound to be in an advanced stage of autoimmune destruction of their β-cells. Moreover, diagnosis of incipient, pre-clinical diabetes by the detection of immunological markers of β-cell autoimmunity can be made only after the onset of the autoimmune process. Therefore, the therapeutic quest is to find a safe, specific and effective way to turn off an autoimmune process that is already well underway.

The present inventors have examined this question before by studying the spontaneous diabetes developing in mice of the NOD strain, which is considered to be a faithful model of human IDDM (23-25). NOD mice develop insulitis around one month of age, which begins as a mild peri-isl t infiltrate and progresses to severe intra-islet inflammation. Hyperglycemia,

which attests to insulin insufficiency, begins in the females in our colony at about three months of age. By six months of age, almost all the female NOD mic have developed severe diabetes and most die in the absence of insulin treatment. Male NOD mice have a lower incidence of disease, for reasons that are not clear. The diabetes of NOD mice has been shown to be caused by autoimmune T cells (26).

T cell reactivity and auto-antibodies to various antigens have been detected in human IDDM patients as well as in NOD mice (27), and it is not clear whether immunity to any single one of the possible target antigens is the primary cause of the disease. Beyond the question of causation is the question of therapy.

It has been demonstrated that the initiation of the autoimmune process in NOD mice can be prevented by subjecting the mice, before the onset of diabetes, to various manipulations such as restricted diet, viral infections, or non-specific stimulation of the immune system (24). NOD diabetes is also preventable by induction of immunological tolerance in pre-diabetic mice to the antigen glutamic acid decarboxylase (GAD) (28, 29).

Anti-idiotypic T cells are T cells that recognize peptides derived from the antigen receptors of other T cells (6). It is thought that anti-idiotypic T cells are involved in regulating the activities of the T cells whose T cell receptor (TCR) peptides they recognize. Autoimmune T cells might be subject to regulation by anti-idiotypic T cells: anti-idiotypic T cells have been detected following intentional T cell vaccination of rodents in the model of experimental autoimmune encephalomyelitis (EAE) (6) or of humans (7) suffering from multiple sclerosis (MS) with autoimmune T cells.

European patent application 261,648 discloses the use of activated T cells specific for an autoimmune disease for the treatment of such disease. The T cells are preferably first pressure treated, subjected to a chemical cross-linking agent and/or subjected to a cytoskeletal disrupting agent in order to improve their immunogenicity. The entire treated

CLAIMS

1. A peptide having at least 7 and preferably about 7-24 amino acid residues, substantially corresponding to a "VDJ" region of the formula

V-D-J

in which "V" includes the dipeptide sequence *A-S*, "D", preferably having 2-5 amino acid residues, includes the dipeptide sequence *L-G*, and "J" includes the tripeptide sequence *N-Q-D*, or a salt or functional derivative thereof.

2. The peptide of claim 1 in which "V" includes the tripeptide sequence *A-S-S*.

3. The peptide of claim 1 in which "D" includes the tripeptide sequence *L-G-G*, the tripeptide sequence *R-L-G* or the pentapeptide sequence *L-G-L-G-A* (residues 4-8 of SEQ ID NO:4).

4. The peptide of any of claims 1-3, wherein the dipeptide sequence *A-S* of "V" is part of the tripeptide adjacent to "D".

5. The peptide of any of claims 1-5, wherein the tripeptide sequence *N-Q-D* of "J" is adjacent to "D".

6. The peptide of any of claims 1-5, wherein "V" comprises a "Vβ" segment which segment includes at least a portion of the C-terminal end of a protein encoded by a Vβ gene.

7. The peptide of claim 6, wherein the C-terminal tripeptide sequence of said Vβ segment includes the dipeptide sequence A-*S*.

8. The peptide of claim 6 in which said segment includes from 1 to about 10 amino acid residues of the C-terminal end of the protein encoded by the Vβ gene.

9. The peptide of claim 6, in which said Vβ gene is selected from the group consisting of Vβ6, Vβ8, Vβ12, and Vβ16.

10. The peptide of any claims 1-3, wherein "J" comprises a "Jβ" segment which segment includes at least a portion of the N-terminal end of a portion encoded by a Jβ gene.

11. The peptide of claim 10, wherein the N-terminal tripeptide sequence of said Jβ segment is *N-Q-D*.

12. The peptide of claim 10 in which said segment includes from 1 to about 10 amino acid residues of the N-terminal end of the protein encoded by the Jβ gene.

13. The peptide of claim 10 in which said Jβ gene is Jβ2.5.

14. A peptide having up to about 24 amino acid residues comprising the sequence *A-S-S-L-G-G-N-Q-D* (residues 47-55 of SEQ ID NO:1).

15. A peptide having up to about 24 amino acid residues comprising the sequence *A-S-R-L-G-N-Q-D* (SEQ ID NO:3).

16. A peptide having up to about 24 amino acid residues comprising the sequence *A-S-S-L-G-L-G-A-N-Q-D* (SEQ ID NO:4).

17. A peptide having up to about 24 amino acid residues comprising the sequence *A-S-S-L-G-A-N-Q-D* (SEQ ID NO:16).

18. A DNA construct comprising a polynucleotide sequence encoding the peptide of any of claims 1-3 or 14-17, or its complement.

19. A pharmaceutical composition comprising a peptide of any of claims 1-3 or 14-17.

20. The pharmaceutical composition of claim 19 which is a vaccine.

21. An agent for detecting the presence of anti-idiotypic T cells involved in the recognition of anti-p277 T cells comprising a peptide of any of claims 1-3 or 14-17.

22. The agent of claim 21 which is conjugated to a detectable label.

23. A conjugate comprising the peptide of any of claims 1-3 or 14-17 and a second molecule.

24. The conjugate of claim 23 in which said second molecule is a polypeptide.

25. The conjugate of claim 23 in which said second molecule is a small organic molecule.

26. A method of modulating anti-idiotypic T cell activity in an individual, said activity associated with the ability of anti-idiotypic T cells to recognize anti-p277 T cells, comprising administering to the individual an amount of a peptide in accordance with any of claims 1-3 or 14-17 effective to regulate the activity of said anti-idiotypic T cells.

27. The method of claim 26 in which said activity is potentiated.